Study on age-related bioaccumulation of some heavy metals in the soft tissue of rock Oyster (*Saccostrea cucullata*) from Laft Port – Qeshm Island, Iran

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Abstract

Heavy metals widely enter into aquatic ecosystems, and cause various environmental problems due to bioaccumulation and biomagnification in food chains. The accumulation of heavy metals in bivalve tissues is affected by a number of intrinsic and extrinsic factors such as physiological conditions, growth, seasonal changes, pH, salinity, temperature, genera and age. The present study investigated the effects of age of the rock oyster *Saccostrea cucullata* on the accumulation of Ni, Cd and Pb in the Laft Port coast located on the Qeshm Island. 200 oysters were collected and their age was determined, then they were classified into four age categories and 15 oysters from each category were selected. Samples were dry digested and the metal concentrations were measured by an ICP-OES instrument. Results revealed that the accumulation of Ni and Pb in one year old oysters (immature) was more than those in mature oysters (two, three and four year old oysters). Significant differences were observed between concentrations of Ni and Pb in mature and immature oysters. The results suggested that aging has a negative effect on bioaccumulation of Ni and Pb in *S. cucullata*; while it has no effect on bioaccumulation of Cd.

Keywords: Bioaccumulation, Age, Rock oyster, Heavy metals, Laft Port–Qeshm Island

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Introduction

Understanding the fundamental principles and rules of water science is necessary for the proper management of water resources (Mirzajani *et al*., 2016). In recent decades, aquatic ecosystems were polluted by persistent pollutants (heavy metals) discharged from agricultural and industrial resources (Hamidian, 2014; Uysal *et al*., 2008). Heavy metals continuously enter into the aquatic ecosystems from anthropogenic resources. They are counted as a serious threat to the food chain due to bioaccumulation, toxicity, long term persistence and aggregation behavior; which result in biodiversity reduction in marine ecosystems (Dixon, 1996; Laimanso *et al*., 1999; Tekin-Ozan and Kir, 2007). Heavy metals can accumulate in the body of aquatic organisms through different ways such as respiration, absorption and ingestion. They have serious effects on aquatic organisms due to their toxicity and bioaccumulation in food chains. The extinction of many species has been reported, because they were not resistant to metal toxicity (Nasehi *et al*., 2012; Zhou *et al*., 2001). The bioaccumulation and magnification of heavy metals, via food chain, can also threaten human health (Agah *et al*., 2009). Although some heavy metals such as zinc, copper and iron are essential in low concentrations for the metabolism of organisms; others such as arsenic, cadmium and nickel are harmful even in low concentrations (Cebrian and Uriz, 2007). Organisms are linked to aquatic ecosystems in different levels of the food chain. Among them, particle feeding organisms play an important role in the food chain of pelagic and benthic zones (Dame, 1972, 1976 and 1996). Heavy metals are absorbed by bivalves directly through breathing in the gills or indirectly through digesting food particles (Clark, 1997). The ability of metal absorption through gills is higher than the gastrointestinal route in bivalves. Bivalves can filter large amounts of water through their gills; therefore they show a great ability to be exposed to various pollutants (Naimo, 1995; Salahshur *et al*., 2014). During the respiration process, a large volume of water passes through the gills and consequently, contaminants are easily absorbed due to high absorption potential of gills (Tinsley, 1979).

Soft tissues of bivalves can be used as bio-indicator of heavy metals in coastal regions. Intrinsic and extrinsic factors affect heavy metal accumulation in tissues of bivalves (Yap *et al*., 2006). Extrinsic factors include physiological conditions, growth, seasonal changes, pH, salinity, temperature, genus and age (Jakimska *et al*., 2011). The purpose of this study is to provide appropriate information to understand physiological strategies of Ni, Cd and Pb accumulation in relation to age of the rock oyster *Saccostrea cucullata*, in the coast of Laft Port located on the Qeshm Island.

Material and methods

The present study investigates the effects of age on the concentration of heavy metals in the rock oyster *S. cucullata* collected from the coastal areas of the Laft Port located on Qeshm Island (26°56′ N, 55°43′ E). In the late summer of 2011, 200 specimens of the oyster (in different sizes) were collected using a chisel and a hammer (Fig. 1).

Mud and sediments of the shells were washed with seawater (Parafkande Haghighi, 2000). The samples were placed in thermal isolation polystyrene bags containing ice and transported into the Environmental Pollution Laboratory at the University of Tehran. In the laboratory, the soft tissues of oysters were dissected by an acid washed plastic knife and located into conical flasks. All glassware used in this study were washed three times with concentrated nitric acid and rinsed three times with distilled water (Einollahi Peer *et al*., 2010). Soft tissues were weighed to the nearest 0.001 g and placed in the oven for 48 hours at a temperature of 110°C and then dry weights were recorded. The dried samples were ashed using a furnace at 450°C for 72 h (the temperature of the furnace was set at 50°C at the start and increased to 450°C in 50°C intervals in an hour). The ashed samples were digested with 10 mL concentrated nitric acid (Merck, Germany) on a hot plate and after acid evaporation the remains were diluted into 25 mL of 1% nitric acid. The concentrations of Cd, Ni and Pb in the solutions were measured by an ICP- OES[1] (GBC Integra XL, Australia) instrument.

The shells are commonly used for age determination. Rings on the shells reflect variation in the growth rate of bivalves which are similar to the hard structures of fish including one light ring in the fast growing season and one thinner dark ring in the cold season.

After cutting the top shell of oyster and dividing umbo by hacksaw, the age of oyster *S. cucullata* was determined by counting the dark and light rings of the shell (Parafkande Haghighi, 2000).

Oysters were classified in four age categories, and then from each category fifteen oysters were selected; a total of 60 soft tissues:

1. 2≥: {2, +1, 1}
2. 3≥: {3, +2}
3. 4≥: {4, +3}
4. 4<: {+5, 5, +4}

Kolmogorov-Smirnov's test was used to determine data normality and homogeneity. General and multiple comparisons were investigated by statistical methods one-way-ANOVA and Duncan's test, respectively. All statistical analyses were performed by SPSS16[2] software.

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[1] Inductively coupled plasma optical emission spectrometry.

[2] Statistical Package for the Social Sciences.

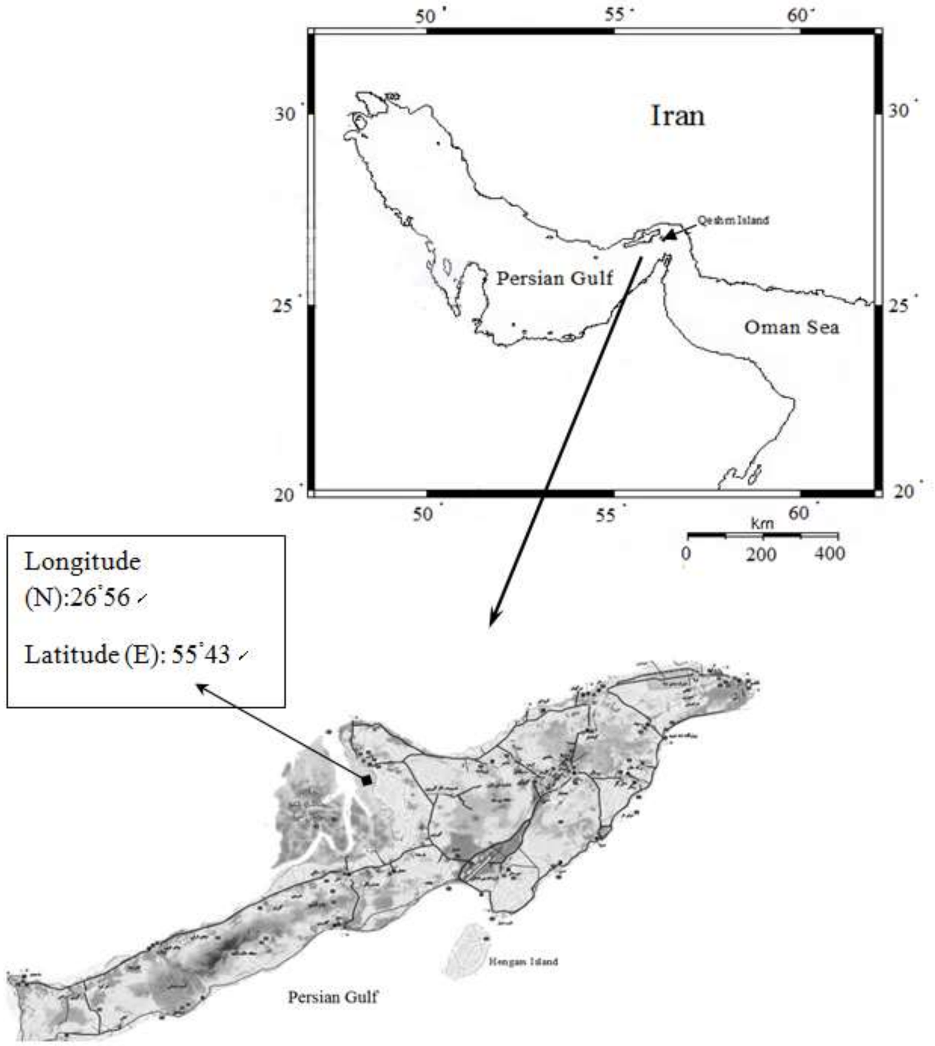

Figure 1: Location of the study area in 2011.

Results

According to the results of morphometric measurements, the age of oysters varied between one and four years. The results of the ANOVA (Table 1) showed significant relationships between age and the concentrations of Pb and Ni ($p<0.01$), but no significant difference was found between age and Cd concentrations in tissues.

Based on the results (Fig. 2), the oysters belonging to the age category one had significantly highest concentrations of Pb and Ni. Two different classes of oysters were recognized based on the effects of age on Pb and Ni concentrations. However, based on Cd concentrations, all oysters were statistically classified in one category.

Table 1: Results of ANOVA: Age effects on metal concentrations (Ni, Cd and Pb).

Factor	Metal	df	F	*p*-value	
Age	Ni	59	10.90	<0.001	**
Age	Cd	59	0.66	0.584	Ns
Age	Pb	59	15.80	0.003	**

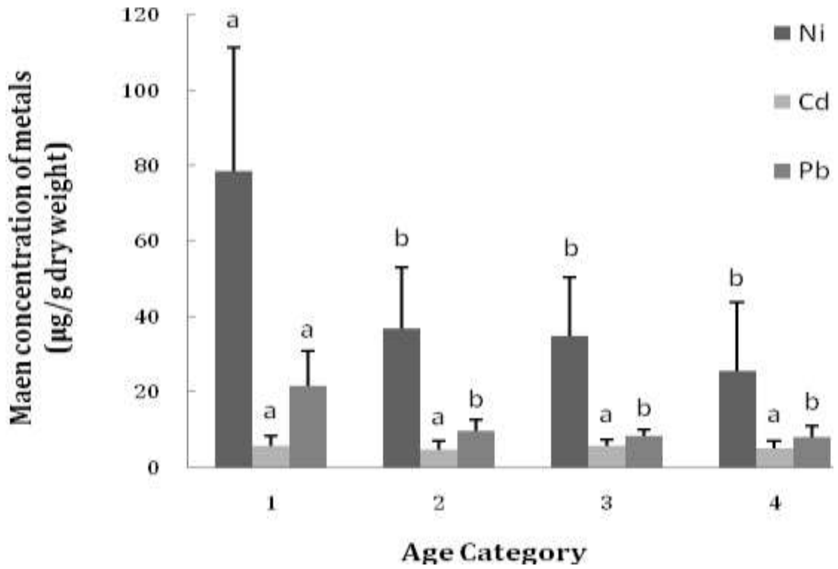

Figure 2: Mean concentration of metals in the soft tissues of oysters (mean + SE) divided into four age categories. Letters on the columns are in relation to the results of Duncan test. Lack of similar letters in each column indicates significant difference at 1% level (a>b).

Discussion

In this research, the effects of age on Cd, Ni and Pb concentrations in soft tissues of oyster *S. cucullata* were investigated. The results indicated that Ni and Pb concentrations in small oysters (class one) were significantly higher than others (classes 2, 3 and 4). Many studies found negative relationships between age and accumulation of Pb and Ni and expressed that the metal concentrations decrease by an increase in age (Farkas, 2002; Yap *et al.*, 2003 and 2009; Rabinson, 2005).

According to Ashjaardalan (2000) and the present study immature oysters have a weight less than two gram and a length between two and three centimeters. Thus, class one is considered as one year old oysters while mature oysters belonged to classes two, three and four. Immature bivalves require a large amount of food and filter feeding due to faster growth rate and higher metabolic rate than mature ones (Savari, 1990). Small bivalves pump higher volumes of water per unit of their body mass compared to adults; therefore, they can be exposed to absorption and elimination of metals (Yap *et al.*, 2009). Mature bivalves accumulate lower concentrations of Ni and Pb than immature bivalves due to factors such as spawning that leads to sudden loss of heavy metal burden and body weight. However, different sizes of bivalve accumulate heavy metals in different concentrations, based on surface to volume ratio and their metabolic requirements (Hedouin *et al*., 2006). Reverse relationship between body size and metal concentration in bivalves shows that a significant part of the metal content is adsorbed. Larger bivalves have smaller surface to volume ratio; thus, the high concentrations of heavy metals are observed in smaller and immature bivalves in comparison with adults (Jones *et al*., 1992).

The results showed that age of oyster has no effect on Cd bioaccumulation and oysters with different ages have statistically similar concentrations of Cd. The findings of this study are in agreement with Rabinson *et al*. (2005), who expressed that age of oyster *Saccostrea glomerata* has no effect on Cd accumulation.

This might be because Cd in oyster tissues might not decrease during the spawning period; therefore mature bivalves have high concentrations of Cd during the spawning period (Zarogian, 1980). Probably, oysters retain Cd in their body and do not excrete it due to similarity with essential metals such as zinc, calcium and copper which are necessary for biological processes such as enzyme function, cell hemostasis and active membrane ion pumps (Holwerda, 1991).

Nonessential metals compete with places in which essential metals are linked to them, for example Cd can compete with Zn for connection to the linking places (due to high tendency to the sulfur ligands) (Fan, 2002). Variation of dissolved calcium concentration can affect Cd absorption (Hamidian and Alavian Petroody, 2014). Similar atomic radius of Cd and Ca may lead to absorption of Cd through Ca channels (Jacobson and Turner, 1980; Hinkle *et al*., 1987; Banaoui *et al*., 2004; Yap *et al*., 2004). Oysters have calcareous shells and they require Ca for shell growth. Due to high concentrations of Cd and/or low Ca concentrations in the surrounding environment, oysters might use Cd instead of Ca. This might be the reason that no significant differences were found between Cd concentrations in mature and immature oysters.

Savari (1990) expressed that the concentration of Cd decreases in oyster *Cerastoderma edule* by an increase in age. Younger oysters have higher concentrations of Cd than older oysters due to physiological differences and different strategies for metabolism (Gavrilovic, 2007; Yap *et al*., 2009). Moreover, Rasmussen *et al*. (2007) stated that the concentration of Cd in *Crassostrea gigas* increases with the increase in age and revealed that most oysters accumulate Cd in the first two years of their life; and after that Cd accumulation gradually increases. Therefore, older oysters have higher concentrations of Cd than younger ones. It can also be due to the reduced ability of older oysters to excrete Cd from their body (Khristoforova, 1989). Variations in Cd accumulation trends can be caused by a number of growth-related changes in physiology, gonad development, food preferences, and MT/ lysosome detoxification systems. Differences in Cd concentrations in different studies may originate from variations in certain intrinsic and extrinsic properties including species, ploidy, habitat, age and weight, season, source of Cd exposure, and metal detoxification systems (Rasmussen *et al*., 2007).

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